

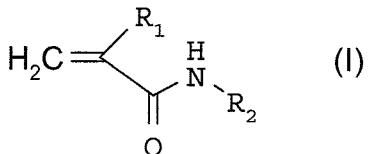
## **AMENDMENTS TO THE CLAIMS:**

This listing of claims will replace all prior versions, and listings, of claims in the application:

## **LISTING OF CLAIMS:**

1. (Currently Amended) A pharmaceutical composition comprising a pharmaceutically acceptable carrier or excipient and, as active ingredient,

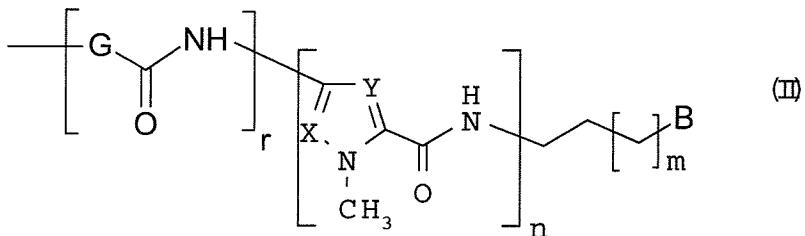
- an acryloyl distamycin derivative of formula (I):



wherein:

$\text{R}_1$  is a bromine or chlorine atom;

$\text{R}_2$  is a group of formula (II)



wherein

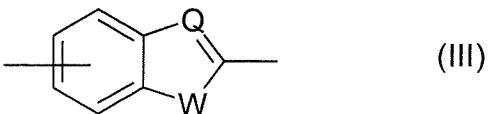
m is an integer from 0 to 2;

n is an integer from 2 to 5;

r is 0 or 1;

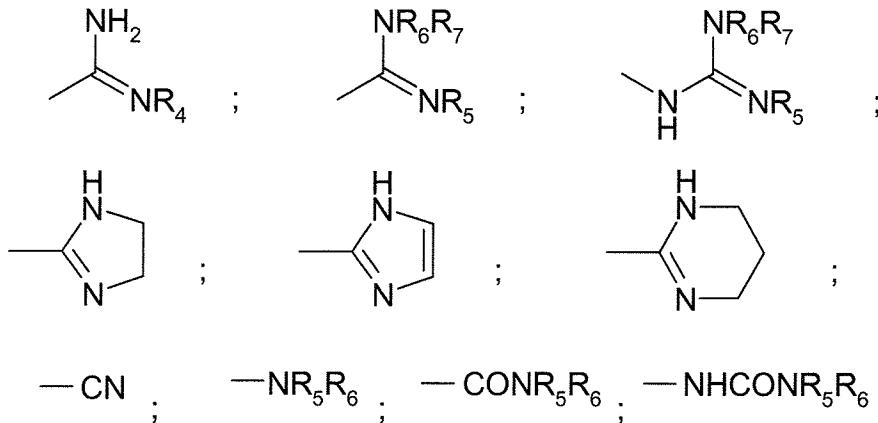
X and Y are, the same or different and independently for each heterocyclic ring, a nitrogen atom or a CH group;

G is phenylene, a 5 or 6 membered saturated or unsaturated heterocyclic ring with from 1 to 3 heteroatoms selected among N, O or S, or it is a group of formula (III) below:



wherein Q is a nitrogen atom or a CH group and W is an oxygen or sulfur atom or it is a group NR<sub>3</sub> wherein R<sub>3</sub> is hydrogen or C<sub>1</sub>-C<sub>4</sub> alkyl;

B is selected from the group consisting of



wherein R<sub>4</sub> is cyano, amino, hydroxy or C<sub>1</sub>-C<sub>4</sub> alkoxy; R<sub>5</sub>, R<sub>6</sub> and R<sub>7</sub>, the same or different, are hydrogen or C<sub>1</sub>-C<sub>4</sub> alkyl; or a pharmaceutically acceptable salt thereof; and

a protein kinase inhibitor; wherein said pharmaceutical composition has a synergistic antineoplastic an antitumoral effect; and wherein said protein kinase inhibitor is selected from the group consisting of STI571(Benzamide, 4-[(4-methyl-1-piperazinyl)methyl]-N-[4-methyl-3-[[4-(3-pyridinyl)-2-pyrimidinyl]amino]phenyl]-), OSI-774(4-Quinazolinamine, N-(3-ethynylphenyl)-6, 7-bis(2-methoxyethoxy)-), PKI 166(Phenol, 4-[4-[[1R]-1-phenylethyl]amino]-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-, EKB-569(2-Butenamide, N-[4-[(3-chloro-4-fluorophenyl)amino]-3-cyano-7-ethoxy-6-quinoliny]-4-(dimethylamino)-(2E)-), GW572016(4-Quinazolinamine, N-[3-chloro-4-[(3-fluorophenyl)methoxyl]phenyl]-6-[5-[[2-(methylsulfonyl)ethyl]amino]methyl]-2-furanyl)-, CEP 2563( $\beta$ -Alanine, L-Iysyl-,[<sup>1</sup>(9S,10S,12R)-2,3,9,10,11,12-hexahydro-10-methoxy-9-methyl-1-oxo-9,12-epoxy-1H-diindolo[1,2,3-fg:3',2',1'-kl]pyrrolo[3,4-I][1,6]benzodiazocin-10-yl]methyl ester, hydrochloride (1:2)), UCN-01(9,13-Epoxy-1H,9H-diindolo[1,2,3-gh:3',2',1'-Im]pyrrolo[3,4-j][1,7]benzodiazonin-1-one,2,3,10,11,12,13-hexahydro-3-hydroxy-10-methoxy-9-methyl-11-(methylamino)-(3R,9S,10R,11R,13R)-), [[G]]CGP 41251(Benzamide, N-[(9S,10R,11R,13R)-2,3,10,11,12,13-hexahydro-10-methoxy-9-methyl-1-oxo-9,13-epoxy-1H,9H-diindolo[1,2,3-gh:3',2',1'-Im]pyrrolo[3,4-j][1,7]benzodiazonin-11-yl]-N-methyl-), Safingol(1,3-Octadecanediol, 2-amino-, (2S, 3S)-), Perifosine(Piperidinium, 4-[[hydroxy(octadecyloxy)phosphinyl]oxy]-1,1-dimethyl-, inner salt), SU 5416(2H-indol-2-one, 3-[(3,5-dimethyl-1H-pyrrol-2-yl)methylene]-1,

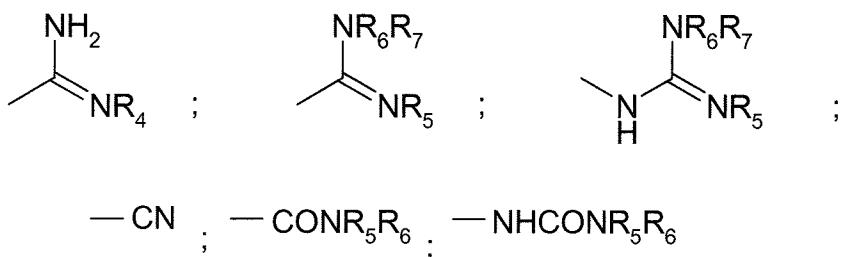
3-dihydro-), CGP 79787(1-Phthalazinamine, N-(4-chlorophenyl)-4-(4-pyridinylmethyl)-), ZD 6474(4-Quinazolinamine, N-(4-bromo-2-fluorophenyl)-6-methoxyl-7-[(1-methyl-4-piperidinyl)methoxyl]-), SU-11248(Butanedioic acid, 2-hydroxy-, (2S)-, compd. with N-[2-(diethylamino)ethyl]-5-[(Z)-(5-fluoror-1,2-dihydro-2-oxo-3H-indol-3-ylidene)methyl]-2,4-dimethyl-1H-pyrrole-3-carboxamide (1:1)), and Flavopiridol(4H-1-Benzopyran-4-one, 2-(2-chlorophenyl)-5,7-dihydroxy-8-[(3S,4R)-3-hydroxy-1-methyl-4-piperidinyl]-); where the antitumoral effect of said pharmaceutical composition is enhanced relative to the additive antitumoral effect of the acryloyl distamycin derivative and the protein kinase inhibitor.

2. (Cancelled)

3. (Previously Presented) The pharmaceutical composition according to claim 1 wherein the protein kinase inhibitor is selected from the group consisting of STI571(Benzamide, 4-[(4-methyl-1-piperazinyl)methyl]-N-[4-methyl-3-[[4-(3-pyridinyl)-2-pyrimidinyl]amino]phenyl]-), OSI-774(4-Quinazolinamine, N-(3-ethynylphenyl)-6, 7-bis(2-methoxyethoxy)-) and SU 5416(2H-indol-2-one, 3-[(3,5-dimethyl-1H-pyrrol-2-yl)methylene]-1, 3-dihydro-).

4. (Cancelled)

5. (Previously Presented) The pharmaceutical composition according to claim 1 comprising an acryloyl distamycin derivative of formula (I) wherein R<sub>1</sub> and R<sub>2</sub> are as defined in claim 1, r is 0, m is 0 or 1, n is 4, X and Y are both CH groups and B is selected from:



wherein R<sub>4</sub> is cyano or hydroxy and R<sub>5</sub>, R<sub>6</sub> and R<sub>7</sub>, the same or different, are hydrogen or C<sub>1</sub>-C<sub>4</sub> alkyl.

6. (Previously Presented) The pharmaceutical composition according to claim 5 comprising an acryloyl distamycin derivative of formula (I) wherein R<sub>1</sub> is bromine, R<sub>2</sub> is a group of formula (II) wherein r and m are 0, n is 4, X and Y are CH, B is a group of formula



wherein R<sub>5</sub>, R<sub>6</sub> and R<sub>7</sub> are hydrogen atoms, optionally in the form of a pharmaceutically acceptable salt.

7. (Previously Presented) The pharmaceutical composition according to claim 1 comprising an acryloyl distamycin derivative, optionally in the form of a pharmaceutically acceptable salt, selected from the group consisting of:

N-(5-{{(5-{{[(2-{{[amino(imino)methyl]amino}ethyl]amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)-4-[(2-bromoacryloyl)amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride;

N-(5-{{(5-{{[(2-{{[amino(imino)methyl]amino}propyl]amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)-4-[(2-bromoacryloyl)amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride;

N-(5-{{(5-{{[(3-amino-3-iminopropyl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)-4-[(2-bromoacryloyl)amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride;

N-(5-{{(5-{{[(3-amino-3-iminopropyl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)-4-[(2-bromoacryloyl)amino]-1-methyl-1H-imidazole-2-carboxamide hydrochloride;

N-(5-{{(5-{{[(3-amino-3-iminopropyl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)-3-[(2-bromoacryloyl)amino]-1-methyl-1H-pyrazole-5-carboxamide hydrochloride;

N-(5-{{(5-{{[(3-amino-3-oxopropyl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)-3-[(2-bromoacryloyl)amino]-1-methyl-1H-pyrazole-5-carboxamide;

N-(5-{{(5-{{[(2-{{[amino(imino)methyl]amino}ethyl]amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)amino}carbonyl}-1-methyl-1H-pyrrol-3-yl)-4-[(2-chloroacryloyl)amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride;

N-(5-{{(5-{{(3-{{[amino(imino)methyl]amino}propyl)amino]carbonyl}-1-methyl-1H-pyrrol-3-yl)amino]carbonyl}-1-methyl-1H-pyrrol-3-yl)-4-[(2-bromoacryloyl)amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride;

N-(5-{{(5-{{(3-amino-3-iminopropyl)amino]carbonyl}-1-methyl-1H-pyrrol-3-yl)amino]carbonyl}-1-methyl-1H-pyrrol-3-yl)-4-[(2-bromoacryloyl)amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride; and

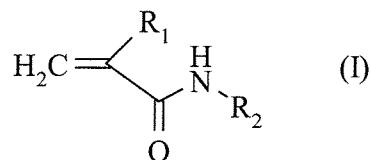
N-{5-[({5-[({3-[(aminocarbonyl)amino]propyl)amino]carbonyl}-1-methyl-1H-pyrrol-3-yl)amino]carbonyl}-1-methyl-1H-pyrrol-3-yl)-4-[(2-bromoacryloyl)amino]-1-methyl-1H-pyrrole-2-carboxamide.

8. (Currently Amended) A pharmaceutical composition comprising a pharmaceutically acceptable carrier or excipient and, as active ingredient,

- N-(5-{{(5-{{(5-{{(2-{{[amino(imino)methyl]amino}ethyl)amino]carbonyl}-1-methyl-1H-pyrrol-3-yl)amino]carbonyl}-1-methyl-1H-pyrrol-3-yl)-4-[(2-bromoacryloyl)amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride (Brostallicin); and

- a protein kinase inhibitor selected from the group consisting of STI571(Benzamide, 4-[(4-methyl-1-piperazinyl)methyl]-N-[4-methyl-3-[[4-(3-pyridinyl)-2-pyrimidinyl]amino]phenyl]-), OSI-774(4-Quinazolinamine, N-(3-ethynylphenyl)-6, 7-bis(2-methoxyethoxy)-), and SU 5416(2H-indol-2-one, 3-[(3,5-dimethyl-1H-pyrrol-2-yl)methylene]-1, 3-dihydro-); wherein the pharmaceutical composition has an antitumoral effect which is enhanced relative to the additive antitumoral effect of the Brostallicin and the protein kinase inhibitor.

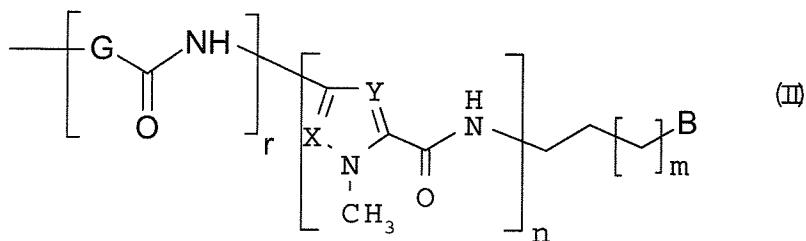
9. (Currently Amended) A product comprising an acryloyl distamycin derivative of formula (I):



wherein:

R<sub>1</sub> is a bromine or chlorine atom;

R<sub>2</sub> is a group of formula (II)



wherein

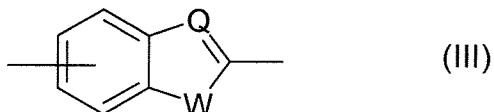
m is an integer from 0 to 2;

n is an integer from 2 to 5;

r is 0 or 1;

X and Y are, the same or different and independently for each heterocyclic ring, a nitrogen atom or a CH group;

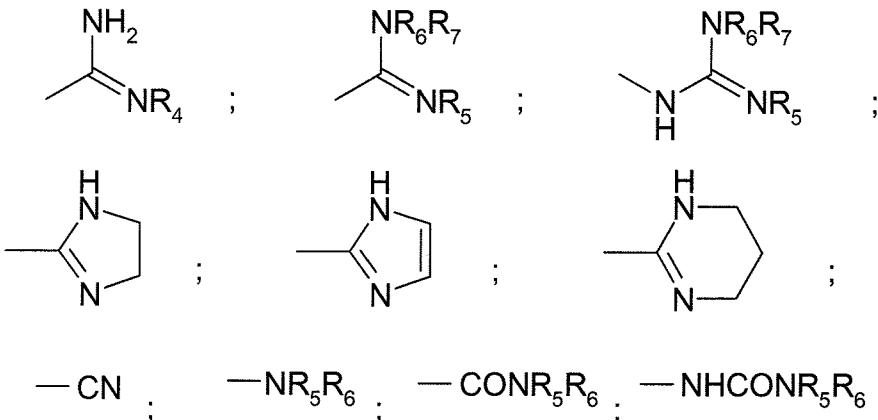
G is phenylene, a 5 or 6 membered saturated or unsaturated heterocyclic ring with from 1 to 3 heteroatoms selected among N, O or S, or it is a group of formula (III) below:



wherein Q is a nitrogen atom or a CH group and W is an oxygen or sulfur atom or it is a group NR<sub>3</sub>;

wherein R<sub>3</sub> is hydrogen or C<sub>1</sub>-C<sub>4</sub> alkyl;

B is selected from the group consisting of



wherein R<sub>4</sub> is cyano, amino, hydroxy or C<sub>1</sub>-C<sub>4</sub> alkoxy; R<sub>5</sub>, R<sub>6</sub> and R<sub>7</sub>, the same or different, are hydrogen or C<sub>1</sub>-C<sub>4</sub> alkyl;

or a pharmaceutically acceptable salt thereof; and

a protein kinase inhibitor, as a preparation where the acryloyl distamycin derivative may be administered simultaneously with the protein kinase inhibitor or, alternatively, both compounds may be administered sequentially in either order in the treatment of tumors; and wherein said protein kinase inhibitor is selected from the group consisting of STI571(Benzamide, 4-[(4-methyl-1-piperazinyl)methyl]-N-[4-methyl-3-[[4-(3-pyridinyl)-2-pyrimidinyl]amino]phenyl]-), OSI-774(4-Quinazolinamine, N-(3-ethynylphenyl)-6, 7-bis(2-methoxyethoxy)-), PKI 166(Phenol, 4-[4-[[1R)-1-phenylethyl]amino]-7H-pyrrolo[2,3-d]pyrimidin-6-yl]-), EKB-569(2-Butenamide, N-[4-[(3-chloro-4-fluorophenyl)amino]-3-cyano-7-ethoxy-6-quinoliny]-4-(dimethylamino)-(2E)-), GW572016(4-Quinazolinamine, N-[3-chloro-4-[(3-fluorophenyl)methoxyl]phenyl]-6-[5-[[2-(methylsulfonyl)ethyl]amino]methyl]-2-furanyl)-), CEP 2563( $\beta$ -Alanine, L-Iysyl-, [(9S,10S,12R)-2,3,9,10,11,12-hexahydro-10-methoxy-9-methyl-1-oxo-9,12-epoxy-1H-diindolo[1,2,3-fg:3',2',1'-kl]pyrrolo[3,4-I][1,6]benzodiazocin-10-yl]methyl ester, hydrochloride (1:2)), UCN-01(9,13-Epoxy-1H,9H-diindolo[1,2,3-gh:3',2',1'-Im]pyrrolo[3,4-j][1,7]benzodiazonin-1-one,2,3,10,11,12,13-hexahydro-3-hydroxy-10-methoxy-9-methyl-11-(methylamino)-(3R,9S,10R,11R,13R)-), [[G]]CGP 41251(Benzamide, N-[(9S,10R,11R,13R)-2,3,10,11,12,13-hexahydro-10-methoxy-9-methyl-1-oxo-9,13-epoxy-1H,9H-diindolo[1,2,3-gh:3',2',1'-Im]pyrrolo[3,4-j][1,7]benzodiazonin-11-yl]-N-methyl-), Safingol(1,3-Octadecanediol, 2-amino-, (2S, 3S)-), Perifosine(Piperidinium, 4-[[hydroxy(octadecyloxy)phosphinyl]oxy]-1,1-dimethyl-, inner salt), SU 5416(2H-indol-2-one, 3-[(3,5-dimethyl-1H-pyrrol-2-yl)methylene]-1, 3-dihydro-), CGP 79787(1-Phthalazinamine, N-(4-chlorophenyl)-4-(4-pyridinylmethyl)-), ZD 6474(4-Quinazolinamine, N-(4-bromo-2-fluorophenyl)-6-methoxyl-7-[(1-methyl-4-piperidinyl)methoxyl]-), SU-11248(Butanedioic acid, 2-hydroxy-, (2S)-, compd. with N-[2-(diethylamino)ethyl]-5-[(Z)-(5-fluoror-1,2-dihydro-2-oxo-3H-indol-3-ylidene)methyl]-2,4-dimethyl-1H-pyrrole-3-carboxamide (1:1)), and Flavopiridol(4H-1-Benzopyran-4-one, 2-(2-chlorophenyl)-5,7-dihydroxy-8-[(3S,4R)-3-hydroxy-1-methyl-4-piperidinyl]-); wherein the product has an antitumoral effect which is enhanced relative to the additive antitumoral effect of the acryloyl distamycin derivative and the protein kinase inhibitor.

10. (Cancelled)

11. (Previously Presented) The product according to claim 9 wherein the protein kinase inhibitor is selected from the group consisting of STI571(Benzamide, 4-[(4-methyl-1-piperazinyl)methyl]-N-[4-methyl-3-[[4-(3-pyridinyl)-2-pyrimidinyl]amino]phenyl]-), OSI-774(4-Quinazolinamine, N-(3-ethynylphenyl)-6, 7-bis(2-methoxyethoxy)-) and SU 5416(2H-indol-2-one, 3-[(3,5-dimethyl-1H-pyrrol-2-yl)methylene]-1, 3-dihydro-).

12. (Cancelled)

13. (Previously Presented) The product according to claim 9 comprising an acryloyl distamycin derivative of formula (I) wherein R<sub>1</sub> is bromine, R<sub>2</sub> is a group of formula (II) wherein r and m are 0, n is 4, X and Y are CH, B is a group of formula



wherein R<sub>5</sub>, R<sub>6</sub> and R<sub>7</sub> are hydrogen, optionally in the form of a pharmaceutically acceptable salt.

14. (Previously Presented) The product according to claim 9 wherein the acryloyl distamycin derivative is selected from the group as defined in claim 7.

15. (Previously Presented) A product comprising the acryloyl distamycin derivative N-[5-[[[5-[[[2-[(aminoiminomethyl)amino]ethyl]amino]carbonyl]-1-methyl-1H-pyrrol-3-yl]amino]carbonyl]-1-methyl-1H-pyrrol-3-yl]-4-[[[4-[(2-bromo-1-oxo-2-propenyl)amino]-1-methyl-1H-pyrrol-2-yl]carbonyl]amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride (Brostallicin), and a protein kinase inhibitor selected from the group consisting of STI571(Benzamide, 4-[(4-methyl-1-piperazinyl)methyl]-N-[4-methyl-3-[[4-(3-pyridinyl)-2-pyrimidinyl]amino]phenyl]-), OSI-774(4-Quinazolinamine, N-(3-ethynylphenyl)-6, 7-bis(2-methoxyethoxy)-) and SU 5416(2H-indol-2-one, 3-[(3,5-dimethyl-1H-pyrrol-2-yl)methylene]-1, 3-dihydro-); as a combined preparation for simultaneous, separate or sequential use in the treatment of tumors.

16.-23. (Cancelled)

24. (Currently Amended) A method of treating a mammal, suffering from a neoplastic disease state, which method comprises administering to said mammal the acryloyl distamycin derivative of formula (I), as defined in claim 1, and a protein kinase inhibitor, in amounts effective to produce a synergistic antineoplastic an antitumoral effect which is enhanced relative to the additive antitumoral effect of the acryloyl distamycin derivative and the protein kinase inhibitor.

25. (Previously Presented) The method according to claim 24 wherein the acryloyl distamycin derivative is N-[5-[[[5-[[[2-[(aminoiminomethyl)amino]ethyl]amino]carbonyl]-1-methyl-1H-pyrrol-3-yl]amino]carbonyl]-1-methyl-1H-pyrrol-3-yl]-4-[[[4-[(2-bromo-1-oxo-2-propenyl)amino]-1-methyl-1H-pyrrol-2-yl[carbonyl]amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride (Brostallicin), and the protein kinase inhibitor is selected from the group consisting of STI571(Benzamide, 4-[(4-methyl-1-piperazinyl)methyl]-N-[4-methyl-3-[(4-(3-pyridinyl)-2-pyrimidinyl]amino]phenyl]-), OSI-774(4-Quinazolinamine, N-(3-ethynylphenyl)-6,7-bis(2-methoxyethoxy)-) and SU 5416(2H-indol-2-one, 3-[(3,5-dimethyl-1H-pyrrol-2-yl)methylene]-1, 3-dihydro-).

26. (Currently Amended) The method for lowering the side effects caused by antineoplastic therapy with an antineoplastic agent, in a mammal in need thereof, the method comprising administering to said mammal a combined preparation comprising a protein kinase inhibitor and an acryloyl distamycin derivative of formula (I), as defined in claim 1, in amounts effective to produce a synergistic antineoplastic an antitumoral effect which is enhanced relative to the additive antitumoral effect of the acryloyl distamycin derivative and the protein kinase inhibitor.

27. (Previously Presented) The method according to claim 26 wherein the acryloyl distamycin derivative is N-[5-[[[5-[[[2-[(aminoiminomethyl)amino]ethyl]amino]carbonyl]-1-methyl-1H-pyrrol-3-yl]amino]carbonyl]-1-methyl-1H-pyrrol-3-yl]-4-[[[4-[(2-bromo-1-oxo-2-propenyl)amino]-1-methyl-1H-pyrrol-2-yl[carbonyl]amino]-1-methyl-1H-pyrrole-2-carboxamide hydrochloride (Brostallicin), and the protein kinase inhibitor is selected from the group

consisting of STI571(Benzamide, 4-[(4-methyl-1-piperazinyl)methyl]-N-[4-methyl-3-[[4-(3-pyridinyl)-2-pyrimidinyl]amino]phenyl]-), OSI-774(4-Quinazolinamine, N-(3-ethynylphenyl)-6,7-bis(2-methoxyethoxy)-) and SU 5416(2H-indol-2-one, 3-[(3,5-dimethyl-1H-pyrrol-2-yl)methylene]-1, 3-dihydro-).

28. (Previously Presented) The method according to claim 24 wherein said disease state is selected from breast, ovary, lung, colon, kidney, stomach, pancreas, liver, melanoma, leukemia and brain tumors.

29. (Currently Amended) A method of treating metastasis suffered by a mammal, which method comprises administering to said mammal the acryloyl distamycin derivative of formula (I), as defined in claim 1, and a protein kinase inhibitor, in amounts effective to produce a synergistic antineoplastic an antitumoral effect which is enhanced relative to the additive antitumoral effect of the acryloyl distamycin derivative and the protein kinase inhibitor.

30. (Currently Amended) A method of treating tumors in a mammal, by inhibition of angiogenesis, which method comprises administering to said mammal the acryloyl distamycin derivative of formula (I), as defined in claim 1, and a protein kinase inhibitor, in amounts effective to produce a synergistic antineoplastic an antitumoral effect which is enhanced relative to the additive antitumoral effect of the acryloyl distamycin derivative and the protein kinase inhibitor.

31. (Previously Presented) The method of treating a mammal according to Claim 24 wherein the mammal is human.

32. (Previously Presented) The method for lowering the side effects according to Claim 26 wherein the mammal is human.

33. (Previously Presented) The method of treating metastasis suffered by a mammal according to Claim 29 wherein the mammal is human.

34. (Previously Presented) The method of treating tumors in a mammal according to Claim 30 wherein the mammal is human.